# Detection of Biological $CO_2$ and 1,3-Pentadiene Using Non-refrigerated Low-Cost MWIR Detectors

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**Abstract** The early detection of spoiling metabolic products in contaminated food is a very important tool to control quality. Some volatile compounds produce unpleasant odours at very low concentrations, making their early detection very challenging. This is the case of 1,3-pentadiene produced by microorganisms through decarboxylation of the preservative sorbate. In this work, we have developed a methodology to use the data produced by a low-cost, compact MWIR (Mid-Wave IR) spectrometry device without moving parts, which is based on a linear array of 128 elements of VPD PbSe coupled to a linear variable filter (LVF) working in the spectral range between 3 and 4.6 μm. This device is able to analyze food headspace gases through dedicated sample presentation setup. This methodology enables the detection of $CO_2$ and the volatile compound 1,3-pentadiene, as compared to synthetic patrons. Data analysis is based on an automated multidimensional dynamic processing of the MWIR spectra. Principal component and discriminant analysis allow segregating between four yeast strains including producers and no producers. The segregation power is accounted as a measure of the discrimination quality.

**Keywords** Gas detector · Sorbate · Food spoilage · Food preservation · Yeast

## Introduction

Yeasts are food spoilage agents of increasing importance which are able to grow in products with low values of pH, water activity and low temperatures. Some species are even able to grow in the presence of preservatives such as low molecular weight weak acids (Loureiro and Querol 1999; Deák 2008). One of the most used preservatives is sorbic acid (E200) and its derived salts (E201, E202 and E203) which are categorized as generally regarded as safe (GRAS). Doses mainly range from 200 to 2000 mg/kg or mg/L, depending on the product (Commission Regulation 2011) as to inhibit or delay the growth of a large number of microorganisms, including yeasts, molds and bacteria.

Foods with added sorbate such as cheeses, soft drinks, jams, margarine, marzipan or bakery products suffer from spoilage which has been referred as plastic smell or hydrocarbons due to the decarboxylation of sorbate into 1,3-pentadiene (Stopforth et al. 2005). Several species of microorganisms have been described as responsible for this decarboxylation: fungi as *Penicillium* sp., *Aspergillus niger* or *Trichoderma* sp. and yeasts such as *Debaryomyces hansenii*, *Saccharomyces cerevisiae* and *Zygosaccharomyces rouxii* (Marth et al. 1966; Finol et al. 1982; Sensidoni et al. 1994; Casas et al. 1996,

1999, 2004; Pinches and Apps 2007; Stratford et al. 2007; Mann and Beuchat 2008).

No adverse effect on human health has been reported at low concentrations of 1,3-pentadiene still odors released during food spoilage generate huge economic losses (Stratford 2006). A clear example of such impact is referred by Gürbüz (2011), who identified the presence of 1,3-pentadiene in 21 supermarket products, containing sorbates: amounts ranging from 5 to 5000 mg/L in foods such as black olives, green olives in brine, honey, bread, cookies, soy sauce, salad and cheese.

Although sensory detection of 1,3-pentadiene is feasible, a chromatographic analysis is needed for a proper identification (Ridgway et al. 2010). Generally, gas chromatography coupled with mass spectrometry is used for identifying and quantifying 1,3-pentadiene in foods (Marth et al. 1966; Liewen and Marth 1985; Casas et al. 1999, 2004; Pinches and Apps 2007; Nieminen et al. 2008; Plumridge et al. 2010; Gürbüz 2011). This time-consuming technique involves qualified staff and its implementation into the food industry is expensive for detecting and rejecting spoiled batches.

A multidimensional characterization of the headspace (GC-analyzed) of fungal cultures has been used, for example, for the identification of species. Methods include the use of principal component analysis of the volatile organic compounds (VOCs) fingerprint, in order to compress the information, together with unsupervised clustering. The aim is to identify functional groups in a process called chemotyping by the authors. This work also indicates that precision greatly increases when the targets are the functional groups (90–99 %) instead of individual species (55–83 %) (Müller et al. 2013).

Mid-IR spectrometers work in 2500–15,000 nm wave spectral region, with two configurations: (a) dispersive, a monochromator-based system which disperses the individual frequencies of radiation and (b) Fourier transform (FT), an interferometer-based system (Ghosh and Jayas 2009). Within technology based on the mid-infrared region of the electromagnetic spectrum, MWIR spectroscopy (Mid-Wave IR, MWIR, 3–5 μm) shows a large potential in the current IR devices market. The main application of MIR spectroscopy is the identification of organic and organometallic molecules in industrial quality control. Due to its multiple applications (gas detection, chemical analysis, industrial monitoring, combustion and flame characterization, food packaging, etc.) and its outstanding performance (good sensitivity, nondestructive method, velocity of response, among others), this technique is open to very diverse fields of application, such as industrial monitoring and control, agriculture, medicine and environmental monitoring. However, the major barrier for its widespread use outside the laboratory: is the lack of affordable specific key elements such a MWIR light sources and low-cost (real uncooled) detectors; recent developments in electro-optical components are helping to overcome such inconveniences (Linares-Herrero et al. 2012).

An industrial application of the detection of VOCs using MWIR has been patented (Cetin and Akers 2011), which includes the combination of visible and several IR range images, with dynamic analysis of video signals. The field domain of the application is the prophylactic detection of leakages in the petrochemical industry where the expected concentration of VOC is relatively high. The know-how generation of the patent can be followed through several papers (Töreyin and Cetin 2008; Erden et al. 2010). Authors remark the necessity of optical methods compared to chemical ones due to their cost-effective and quick response and highlight the interest of IR sensors due to the existence of strong absorption bands of VOCs in such range.

The objective of this paper is to test the feasibility of non-destructive detection of biological 1,3-pentadiene and $CO_2$ by means of non-refrigerated low-cost MWIR detectors, as a prospective for its use in the Agro-food industry. More precisely, this work tests the ability of such device to detect (and quantify) 1,3-pentadiene gas concentration released by microorganism such as yeasts degrading food in which potassium sorbate is used as a preservative.

## Materials and Methods

### MWIR Spectrometer

In this paper, a new MWIR spectrometry device is used based on linear array of 128 elements of vapor phase deposit (VPD) PbSe coupled to a linear variable filter (LVF) working in the spectral range between 3 and 4.6 μm. This compact device, with no moving parts, is based on a rugged and affordable detector and is suitable to be used in applications which demand high sensitivity, good spectral discrimination, reliability and compactness and where an alternative to the traditional scanning instrument is desired.

The spectroscopy equipment is composed by an IR source and a LX-CORE-S-LVF device (bottom of Fig. 1). The IR source used is a SiC glowbar lamp from Hawkeye Technologies coupled to an aluminum reflector which allow to open and collimate the light beam. The LX-CORE-S-LVF device is a compact infrared spectrometer composed by: an electronic module for LUXELL 128 FPA with LVF (linear variable filter) which includes a FPA detector (+LVF 3.0–4.6 μm), an electronic reading card, an USB communication interface and a Software Suite for acquiring and visualizing spectral data specifically developed by NIT for LUXELL 128-LVF FPA. The focal plane arrays (FPA) detector consists of a linear set of detectors (128 pixels) sensitive in the middle infrared wavelength range (MWIR): 1 and 5 μm. It works at room temperature and does not need any cooling system which reduces its cost (the total price of LUXELL 128-LVF spectrometer is under 10000 €). The system allows dynamic

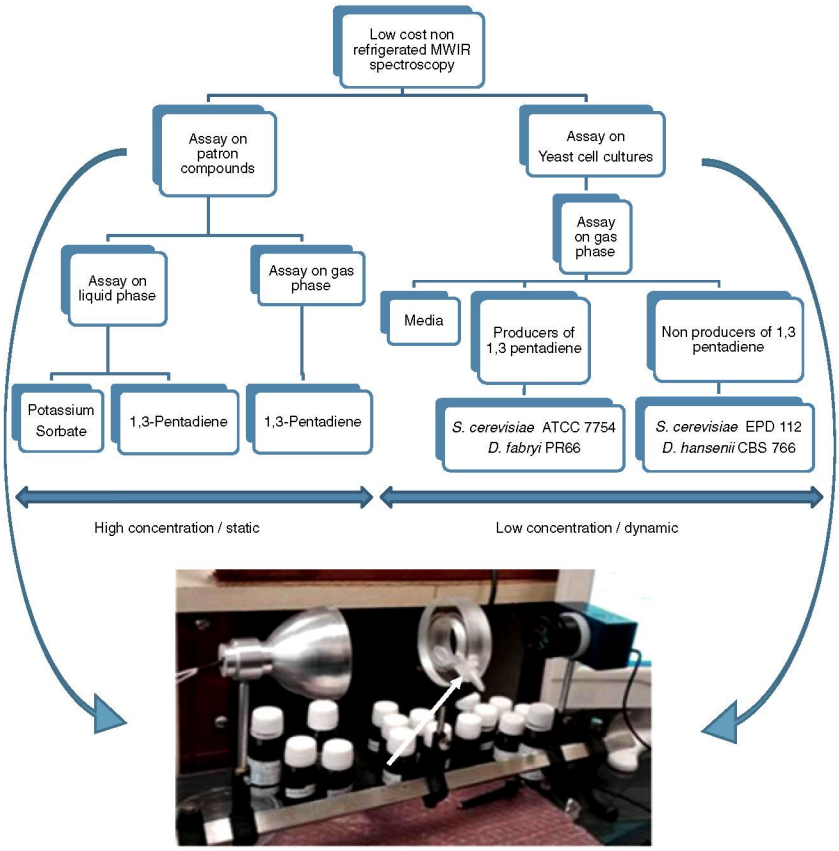


**Fig. 1** Experimental design which covers tests on patron compounds (liquid and gas phases), as well as on yeast cell cultures. At the *bottom* of the figure appears the experimental setup to confine the gas during the spectra measuring. On the *left* is the IR source and on the right the detector. The IR source used is a SiC glowbar lamp coupled to an aluminum reflector which allows to open and collimate the light beam. The LX-CORE-S-LVF device is a compact infrared spectrometer composed by: an electronic module for LUXELL 128 FPA with LVF (linear variable filter) which includes a FPA detector (+LVF 3.0–4.6 μm) with a linear set of detectors (128 pixels). In the middle of these parts, there is a designed chamber to confine the gas. The *arrow* indicates the place to connect the samples to the chamber

recording of data at variable rate, in our case 360 frames per second.

## Experimental Design

Figure 1 summarizes the experimental design used in this work, which covers tests on patron compounds (liquid and gas phases), as well as on yeast cell cultures. Tests on patrons are characterized by a relatively high concentration of volatile organic chemicals (VOCs) in the headspace, and thus, the absorbance bands can readily be identified in isolated spectrum (static measurements). On the other hand, the headspace generated by yeast cells has low VOCs concentration (Casas et al. 2004; Stratford et al. 2007), and therefore, the absorbance peaks are not well resolved. In such cases as this, the feasibility of recording dynamic measurements of the diffusion of VOCs through the measuring chamber is fundamental.

## Assay on Patron Compounds

Patron compounds were tested in liquid and gas phases.

### *Assay on Liquid Phase Compounds*

The measurement in liquid phase was undertaken as follows: First, an empty sapphire wafer's measurement was taken as a blank. Next, a quantity of the solution sufficient to cover the wafer surface was placed between two of them, ranging from lowest dilution to the highest concentration. Three replicates of each concentration were used.

The study started with the preparation of calibration curves using 1,3-pentadiene (Sigma-Aldrich, Wisconsin, USA) at eight concentrations with water, ranging from 0.25 to 700 mg/L. To validate the specificity of the device, some calibration curves were also carried out for potassium sorbate (Scharlau, Barcelona, Spain), ranging from 0.01 to 10 g/L.

### *Assay on Gas Phase Compounds*

An experimental setup was designed to confine the gas between the two wafers in a chamber of 32.26 $cm^3$ volume (see Fig. 1). The distance between light source and detector was selected as to assure a homogeneous illumination of the detector.

For patron compounds, 1 ml of final volume of each concentration of 1,3-pentadiene (using eppendorf tubes, see Fig. 1) was left to volatilize about 1 min, and the measurements were recorded once stabilization was reached (static measurements). The aim of this phase of the experiment was to check the measurements reproducibility (two replicates) and repeatability (3 repetitions per replicate).

## Yeast Cell Cultures

As pentadiene production is a property of the strain, not of the species, assays on yeast cell cultures included: the sterile medium as control, and yeast cultures of four strains. *S. cerevisiae* was represented by two strains: ATCC 7754 as producer, and EPD 1.1.2. as non-producer. *Debaryomyces fabryi* by PR66 (producer) and *D. hansenii* by CBS 766 (non-producer).

The strains used in this work were grown in YM-Broth (0.5 % (*w*/*v*) yeast extract (Difco Laboratories, Detroit, Mitch, USA), 0.3 % (*w*/*v*) proteose-peptone no. 3 (Difco), 0.3 % (*w*/*v*) malt extract (Difco) and 1 % (*w*/*v*) glucose (Panreac Quimica S.A., Barcelona, Spain) plus 0.75 g/L of potassium sorbate at 28 °C for 4 days before the measurement. The inocula had been previously grown without sorbate. At least two replicates for each condition were grown in separated flasks (25 ml) inoculated into two different final volumes of culture media 9 and 18 ml. The flask or tube (25 ml) had a hermetic cap with a faucet to open or close the gas outlet to the experimental setup for gas compounds described above. A total of 21 yeast culture samples and medium were analyzed (5 and 16 corresponding to 9 and 18 ml broth volume, respectively).

For the detection of 1,3-pentadiene in yeast cultures, the same MWIR experimental setup was used recording the spectra (360 frames per second) while the VOCs diffused through the sampling chamber. The test lasted in all cases 370 s. The test was undertaken in three stages: loading (connection of the tube to the measurement capsule), disconnection (disconnection the tube from the capsule) and emptying (time interval over which the capsule was left open for the gas go out through the outlet by diffusion).

## Spectra Analysis

Figure 2 summarizes the procedures used for spectra analysis. All spectra obtained either from liquid or gas phase were processed using Savisky-Golay algorithm to achieve smooth spectra and to compute first derivative. Using the first derivate of the spectrum, three processes were performed: (i) bandwidth identification and comparison of patron compounds with regard to yeast cell cultures, (ii) non-supervised analysis of principal component followed by a non-linear fitting of the first order response of gas diffusion in the experimental setup and ANOVA analysis and clustering and (iii) supervised discriminant analysis and optimization of dynamic gas sampling.

### *Bandwidth Identification*

In an MWIR spectrum, an absorption band is characterized by the presence of an intense decrease in transmittance. The initial and final points of the absorption bands can easily be detected in the first derivative spectra as local minimum and maximum. We consider this procedure more convenient for addressing absorption bands than the traditional definition: wavelength range corresponding to half of the maximum absorption height.

Bandwidths of the spectral curves corresponding to the liquid and gas phase of standard 1,3-pentadiene, as well as the headspace of several yeasts cultures, were calculated using a dedicated MATLAB script designed for computing the bandwidth from the first derivative of the spectral curves.

### *Non-supervised Analysis*

This part of the study gathers the application and transference of MWIR methodology developed for patron compounds, towards the detection of volatile compounds produced by yeast, that characterize food spoilage.

As mentioned before, the 1,3-pentadiene quantity produced by the yeasts is very low compared to the previous patron tests. For this reason, the dynamics of the produced gases were dedicatedly recorded (360 frames/s) as stated in the methodology. In order to visualize the differences between the samples, accurate triggering was set of faucet opening (60 s) and closing (360 s), as well as cleaning (370 s).

The analysis procedure consisted of extracting the intervals of interest (O_MWIR Spectra) from the data files (60–370 s; 103.334 spectra per sample, see Fig. 3) and computing the smooth and first derivative spectra (S_D MWIR Spectra). The intervals extracted were then concatenated (above 2 million spectra) following the experimental order so as to later apply a non-supervised principal component analysis.

The PC scores of each dynamic sample were fit considering a first-order response of gas diffusion in the experimental setup $[a \cdot (1 - e^{(-t/b)}]$, where $a$ is the stabilization signal or the signal level (intensity counts, i.c.), $t$ is the sampling time (ms) and $b$ is the time response or time interval between the initial step and 63 % of final signal of the spectra of each sample (ms). As a result, a stabilization signal, $a$, and time response, $b$, were derived for each sample and principal component. ANOVA analysis was used to test the significance of both volume: 9 and 18 ml.

The samples containing 18 ml of broth were grouped using the Euclidean distance based on the signal stabilization values ($a_i$) of PC2, PC3 and PC4 ($i$=2, 3 and 4 where $i$ refers to the principal component data, PC2, PC3 and PC4, respectively).

### *Supervised Discriminant Analysis*

A supervised analysis of the first derivative spectra (128 variables from 3 to 4.6 μm) was carried out in parallel using an average ($n$=2000, 5.5 s) corresponding to the end of the dynamic signal (370 s) for the 18 ml broth volume (16 samples). Since discrimination is performed among three classes

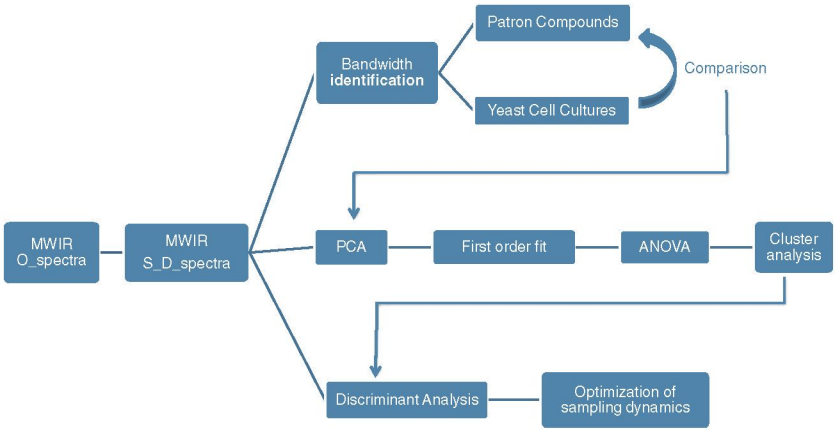


**Fig. 2** Summary of procedures used for spectra analysis. For a detail explanation, see section of "Spectra analysis" in materials and methods

(media, producers and non-pentadiene producers), only 2 canonical variables are needed. As internal validation, the 2000×16 individual spectrums are projected in the canonical plane, for increasing backward sampling time: 370, 367, 362.5, 355, 340, 325 and 280 s. In order to assess the dynamics of segregation in the discriminant analysis during online testing, the area of the triangle, defined by the centroids of the classes in the canonical plane, is used for selected sampling times (seven steps from 60 to 370 s). The area is defined in the space of the discriminant canonical variables. Moreover, the magnitude of such area as compared to the maximum value is computed as a measure of discrimination power.

## Results

### Comparison of Wavelength Absorption Bands

Figure 4 shows the MWIR spectra on liquid phase for increasing concentrations of potassium sorbate (a) and corresponding first derivative (b). The center of the absorption band is highlighted in the figure (3.4–3.45 μm) and ranges from 3.2 to 3.45 μm.

Table 1 indicates the center of the absorption band for liquid and gas experiments. The absorption band corresponding to 1,3-pentadiene seems to be fairly reproducible for liquid and gases phases. Figure 5 shows the MWIR spectra of various concentrations of liquid 1,3-pentadiene (a) compared to MWIR spectra of yeast cultures headspace (b). As expected, for pure compounds, a single absorption band is found (highlighted in red), while two have been addressed for yeast headspace: 1,3-pentadiene (3.2-3.4 μm) and $CO_2$ (4.3 μm). Table 1 also indicates that 1,3-pentadiene in liquid phase is rather overlapped with potassium sorbate. As potassium sorbate has a negligible volatility, a reliable detection of 1,3-pentadiene can only be done in gas phases.

The signal level corresponding to 1,3-pentadiene in yeast culture headspace was much lower (60 i.c in Fig. 5) than that obtained in the liquid phase (170 i.c). This prompted us to develop a dynamic analysis of the spectra (see Fig. 3 as an example).

### Analysis of Dynamic MWIR Gas Spectra

#### *Principal Component Analysis*

A principal component analysis was performed on the first derivative spectra computed by means of Savisky-Golay algorithm ($10^4$ spectra per sample and 21 samples, that is above 2 million spectra). The variance retained with four components reaches 97.2 %. The second principal component (PC2, 31.7 % variance) shows a very neat fingerprint (see Fig. 6a) which has been ascribed to $CO_2$ according to the PC loadings (maximum contribution corresponds to 4.14–4.27 μm). PC3 (Fig. 6b) and PC4 (5.8 and 2.3 % of variance) also provide a consistent dynamic response, with loadings

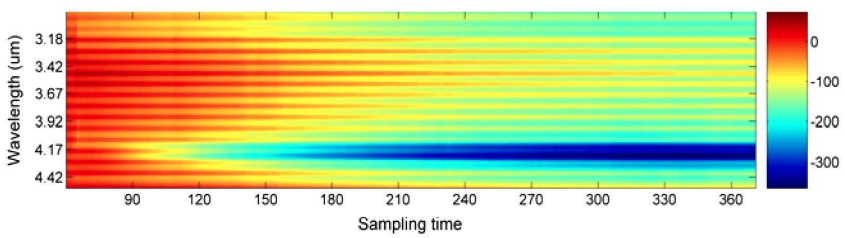


**Fig. 3** Dynamics of spectra acquisition from opening to cleaning in a sample of yeast culture (in color in the online version). The *color scale* show the signal level (intensity counts, the bigger the bluer) at each wavelengths. The two absorption bands appear in this sample, visualize by a decrease in signal (*bluer*)

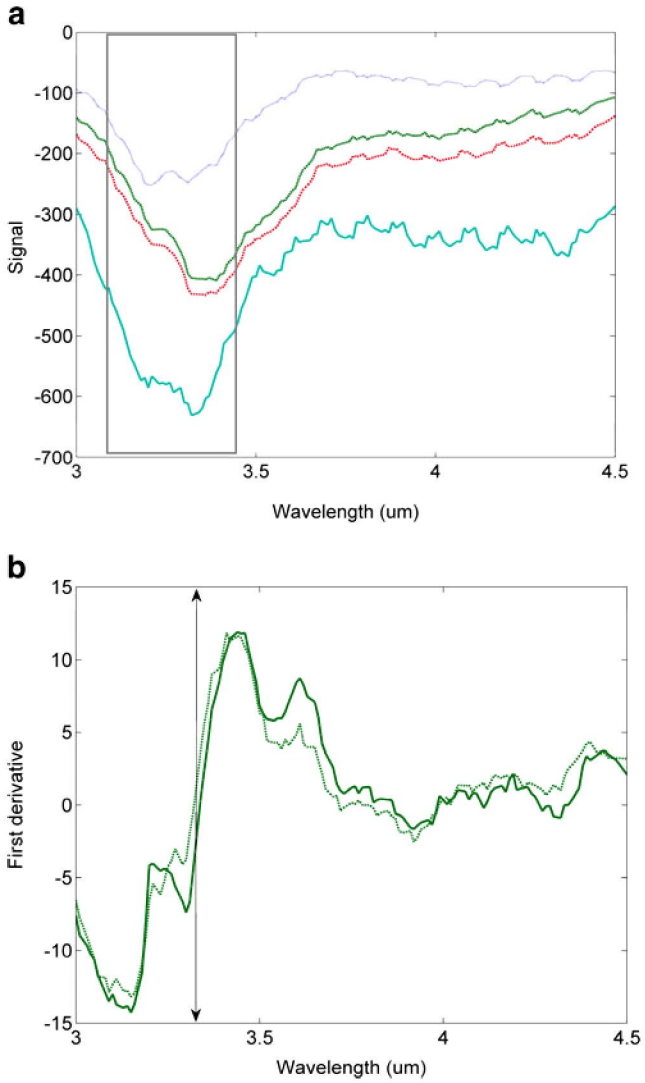


**Fig. 4** MWIR (signal measured as intensity counts) spectra for increasing concentrations of potassium sorbate (0.01, 0.1, 0.5 and 5 g/L) (**a**) and first derivative spectra corresponding to 1,3-pentadiene (dotted line) and potassium Sorbate (continuous line) (**b**). Both are made with liquid phase compounds. The *rectangle* indicates the range of absorption bands characterized by an intense decrease in transmittance (3.2–3.45 μm). The *arrow* shows the center of the absorption band (3.4–3.45 μm)

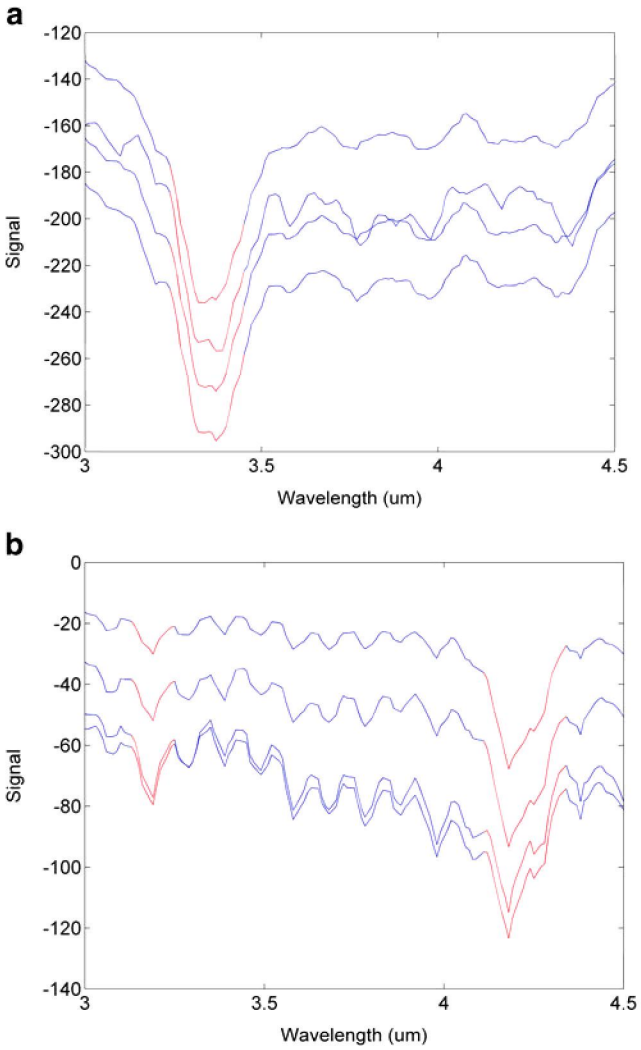


**Fig. 5** MWIR spectra of various concentrations of liquid 1,3-pentadiene (**a**) compared to MWIR spectra of cultures of *D. fabryi* PR66 headspace (**b**). For pure 1,3-pentadiene, a big and single absorption band is detected (*red*), while in yeast culture headspace two bands (*red*) are detected: 3.2–3.4 and 4.3 μm for 1,3-pentadiene and $CO_2$, respectively

pointing to wavelength bands related to that of pentadiene as indicated in Table 1 (3.2–3.4 μm).

### *First order Fit of PCs Time Fingerprint*

The dynamic behavior of PC2, PC3 and PC4 was adjusted to a first-order time response by means of a non-linear fit (above $10^4$ data per sample). The adjusted parameters

**Table 1** Centre of the absorption (μm) band for liquid and gas experiments

| Phase | Compound | Centre of absorption band 1 (μm) ascribed to 1,3-pentadiene | Centre of absorption band 2 (μm) ascribed to $CO_2$ |
|---|---|---|---|
| Liquid | 1,3-Pentadiene | 3.44 | – |
| | Potassium sorbate | 3.45 | – |
| Gas | 1,3-Pentadiene | 3.46 | – |
| | *D. fabryi* PR66 (R. 1) | 3.23 | 4.30 |
| | *D. fabryi* PR66 (R. 2) | 3.31 | 4.33 |
| | *D. fabryi* PR66 (R. 3) | 3.32 | 4.32 |

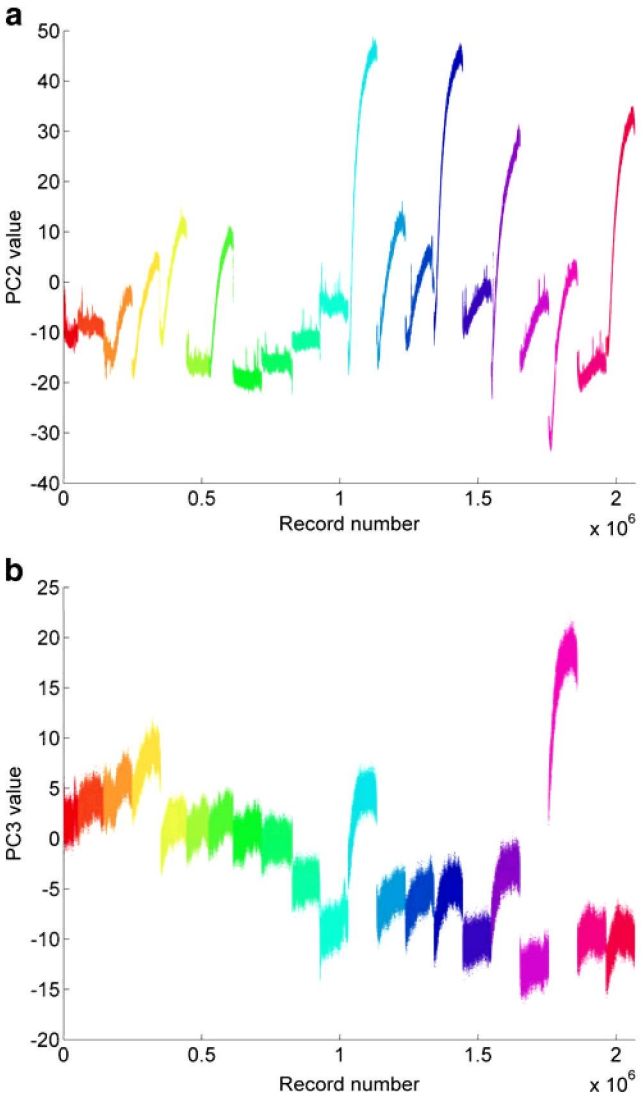


**Fig. 6** **a** PC2 and **b** PC3 scores of the spectra gathered in the 21 dynamic analyses (each sample in different colour with approximately 100,000 spectra per sample). The response in PC2 and PC3 has been ascribed mainly to $CO_2$ (**a**) and 1,3-pentadiene (**b**) respectively

were in all cases the stabilization signal ($a_i$, intensity counts) and the time response ($b_i$, ms), where $i$ refers to the principal component data used for fitting ($i$=2, 3 and 4, PC2, PC3 and PC4, respectively). As expected, the highest signal was found for PC2 (68.18) compared to PC3 and PC4 (14.95 and 17.57, respectively). Table 2 provides the range of such parameters for goodness of fit $r^2$ above 0.85. All maximum response times remain within the acquisition time (6.17 min, 370,000 ms); note that according to a first-order response model, $b_i$ corresponds to the period required to reach 63 % of stabilization signal. The data shown in Table 2 confirmed that the spectra contained relevant information regarding the headspace composition, information which changed in time before stabilizing.

**Table 2** Parameters for first order time response for the dynamic behaviour of PC2, PC3 and PC4

| | PC2 | | PC3 | | PC4 | |
|---|---|---|---|---|---|---|
| | $a_2$ (i.c.) | $b_2$(min) | $a_3$ (i.c.) | $b_3$(min) | $a_4$ (i.c.) | $b_4$(min) |
| Minimum | 17.59 | 1.74 | 1.20 | 0.44 | 1.69 | 0.81 |
| Maximum | 68.18 | 4.99 | 14.95 | 3.60 | 17.57 | 6.59 |

*i.c.* intensity counts

*Anova on the Effect of Yeast Biomass*

Analysis of variance was performed on $a_i$ and $b_i$ in order to check the effect of the broth volume (yeast biomass) used for growing the yeast. Broth volume had a statistically significant effect (<1 %) for $a_2$ and $b_2$ (F=10.95 and 4.39, respectively). Using 18 ml allowed higher signal stabilization (2 times higher signal from 18.8 to 41.2) and 41.7 % decrease in response time compared to 9 ml (5.3 to 3.09 min on average). These results show the positive correlation between the amount of gas produced and the total amount of yeast biomass present, demonstrating its metabolic origin.

*Cluster Analysis*

Taking into account the large effect of broth volume in the signal, it was decided to continue the analysis with those samples exhibiting highest response, that is, to say, 18 ml broth (16 samples). An unsupervised cluster analysis was performed based on $a_2$, $a_3$ and $a_4$ following the ward method and the Euclidean distance. Figure 7 shows the aggregation tree. First of all, it divides the samples according to their $CO_2$ production, in highest and less or non-producers of $CO_2$. Inside this division, samples corresponding to the media are all grouped within the same cluster, and samples corresponding to pentadiene producing yeasts are consistently grouped inside the main two divisions. It was then concluded that the dynamic spectra provided enough information, and that it would be of interest in a further stage to analyze if a short MWIR snapshot (128 wavelengths) would be representative for the assessment.

*Supervised Discriminant Analysis*

Figure 8 shows the projection in the canonical plane of the 16 average spectra (squares), as well as the whole set of internal validation (16×2000). A very neat segregation of all three culture groups of data could be observed: media (blue), pentadiene producers (green) and non-pentadiene producers (red).

*Optimization of Dynamic Analysis*

The segregation power of the groups (pentadiene producers, non-producers and media) for increasing backward sampling

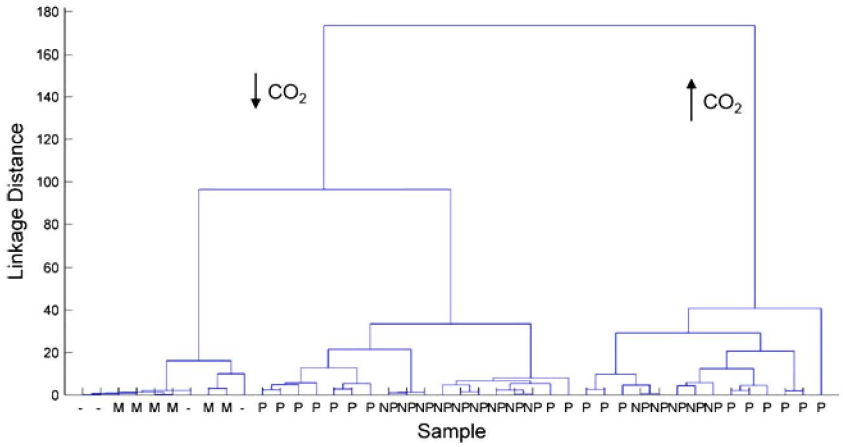


**Fig. 7** Cluster analysis of 18 ml yeast cells cultures (producers of 1,3-Pentadiene, *P*, and non-producers, *NP*) and media (*M*) based on $a_2$, $a_3$ and $a_4$ following the ward method and the Euclidean distance

time, increases from 55 % at 280 s until the maximum segregation power (100 %) which is achieved for a sampling time of 6 min (360 s) where enough time has just passed to diffuse the gas into the chamber.

## Discussion

The early detection of spoiling metabolic products in contaminated food is a very important tool to control its quality (Loureiro and Querol 1999). The isolation and quantification of the contaminating microorganisms are not always useful because the spoiling capacity is not a property of the species identified but of the strain (Wrent et al. 2010). For these reasons, four different strains, belonging to three species of yeasts, with different capacities to produce $CO_2$ and 1,3-pentadiene have been used. In this work, we have developed a methodology to use the data produced by a MWIR spectrometry device able to analyze food headspaces. This methodology enable the detection of $CO_2$ and the volatile compound 1,3-pentadiene as well as the discrimination between high and low producers.

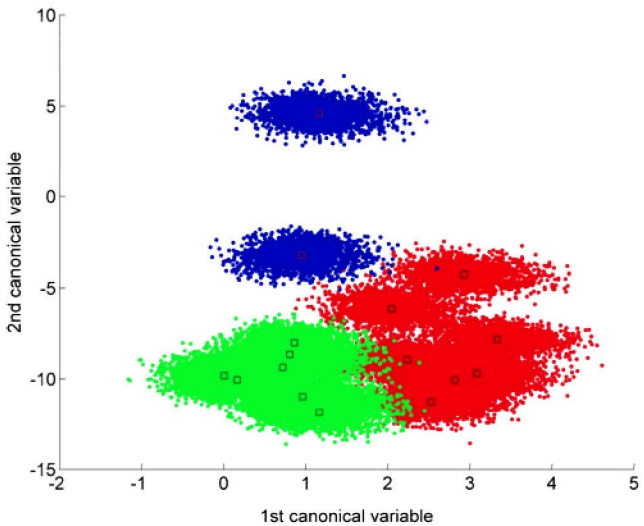


**Fig. 8** Results of supervised discriminant analysis of yeast cultures: calibration is performed on the average spectra (*black squares*) while internal validation is achieved by projecting the individual spectrums (*dots*) (in colour in the online version): *blue dots*: media; *red dots*: non-pentadiene producers; *green dots*: pentadiene producers

Several studies using MWIR spectrometers have described the detection of $CO_2$, for example, Scharf et al. in 2009, who used a micromachined spectrometer based on FT-IR with MIR optical fibers to detect and quantify greenhouse gases such as $CO_2$. In this study, the $CO_2$ had two absorption bands in 2.67 and 4.25 μm. In our study, a single band for $CO_2$ around 4.30 μm is found by our device as expected (the former being out of the LVF range). Although the amounts of $CO_2$ are not comparable, the same device or an optical fiber has been used to analyze the dynamics and concentration of the exhausted $CO_2$ by aircraft turbojet and in the exhaust system of a motor vehicle (Linares-Herrero et al. 2013; Mulrooney et al. 2007). This study confirms the wide applicability of this type of measures based on mid-infrared to detect the production of $CO_2$ in industries where this detection is required, for example, modified atmosphere packaging or fermentation processes.

In our study, we conducted a principal components analysis from the first derivative spectra of the samples. In many studies, this type of analysis is used to reduce the range of the data in new and orthogonal variables that explain the differences in the sample values (scores) (Karoui et al. 2010). In our case, the graphical display of the dynamic behavior of PC2, PC3 and PC4 scores revealed a first-order time response pattern in PC2 related to $CO_2$ and in PC3 and PC4 in the case of 1,3-pentadiene production. Also, Sinelli et al. (2008) reported the use of the NIR and MIR to detect different kinds of flours, associating them with curves obtained by plotting PC1 and PC2 scores versus time. In our case, ANOVA analysis showed a significant effect of the culture volume in the first order time response parameters, $a_i$ and $b_i$. This fact might cause differences when measuring volatile compounds in real food samples.

Regarding the production of odors and flavors in food, there is a list of over 40 types of odors (for a review, see Ridgway et al. 2010). The main cause of most of them is

microbial action. As already described at the introduction, several species of microorganisms have been characterized as 1,3-pentadiene producers from the preservative sorbate decarboxylation. This odour has been described as kerosene, paraffin or plastic odor, and it is easily recognizable in low amounts. The device tested is able to distinguish different concentrations of potassium sorbate in aqueous solution. Absorption bands corresponding to 1,3-pentadiene and potassium sorbate are approximately in the same wavelength, which does not cause any trouble in the gas detection because of the non-volatility of the preservative. One advantage of the MWIR spectrophotometer is that it can detect the low concentration of pentadiene produced by the yeast cultures based on 3.2–3.4 absorption band, including levels of pure pentadiene lower than the sensory threshold value, that has been established on 2.5 ml/l in 10 % brine and 4 mg/kg in cheese (Saxby 1993). The amount of 1,3-pentadiene produced by yeast species such as *D. hansenii* and *Z. rouxii* has been previously quantified using GC/MS and it did not exceed 3 mg/L after 4 days of incubation at 28 °C (Casas et al. 2004) from an initial concentration of 1 g/L or potassium sorbate. A similar percentage of decarboxylation has been detected in *S. cerevisiae* (Stratford et al. 2007). In the study of Gürbüz (2011), food products with up to 5000 mg/L were detected, possibly due to fungal action, which are able to convert 100 % of sorbate (Plumridge et al. 2010) into pentadiene as compared with yeast decarboxylation which amounts no more than 1 % of conversion into pentadiene. For this reason, the development of sensitive methods, as the one described here, is very important when the spoiling agents are yeasts.

Carrying out the test with a higher number of samples from different species (1,3-pentadiene producer and non-producer yeasts) is ongoing to validate the results obtained. The fastest way to reach an industrial application would be by means of a discriminant analysis of the spectra samples, together with the expert evaluation.

Anyway, in order to include this setup for a real application, the first step is to test the interferences due to the plastic materials of food container since organic compounds may have an absorbance band in the same range than 1,3-pentadiene. In such a case, it may be advised the use of any type of low-cost window in the food container, for sampling. It would be better searching for a solution that does not require driving out the headspace of the container, in order to keep a non-destructive procedure.

## Conclusions

This study shows a first approximation of the use of a low-cost and compact MWIR (Mid-Wave IR) spectrometry device to analyze the headspaces of yeast cultures, which works in the spectral range between 3 and 4.6 μm. The gas spectra obtained during a dynamic recording (360 Hz) in yeast cultures is released into an experimental setup (designed to confine the gas) and processed by several multidimensional techniques: principal components and discriminant analysis. The results show that this device with the corresponding data analysis is an easy and low-cost approach to differentiate samples with $CO_2$ and/or 1,3-pentadiene, compounds released by spoiling yeast in foods. The former responsible for swollen containers, while the latter leads to unpleasant odors (lose of commercial value).

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**Compliance with Ethical Standards**

**Conflict of Interest** Eva Mª Rivas declares that she has no conflict of interest. María Maldonado declares that she has no conflict of interest. Belén Diezma declares that she has no conflict of interest. Petra Wrent declares that she has no conflict of interest. José M. Peinado declares that he has no conflict of interest. María-Isabel de Silóniz declares that she has no conflict of interest. Germán Vergara declares that he has no conflict of interest. Javier García-Hierro declares that he has no conflict of interest. José Ignacio Robla declares that he has no conflict of interest. Pilar Barreiro declares that she has no conflict of interest.

**Ethical Approval** This article does not contain any studies with human participants or animals performed by any of the authors.

**Informed Consent** Not applicable.

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